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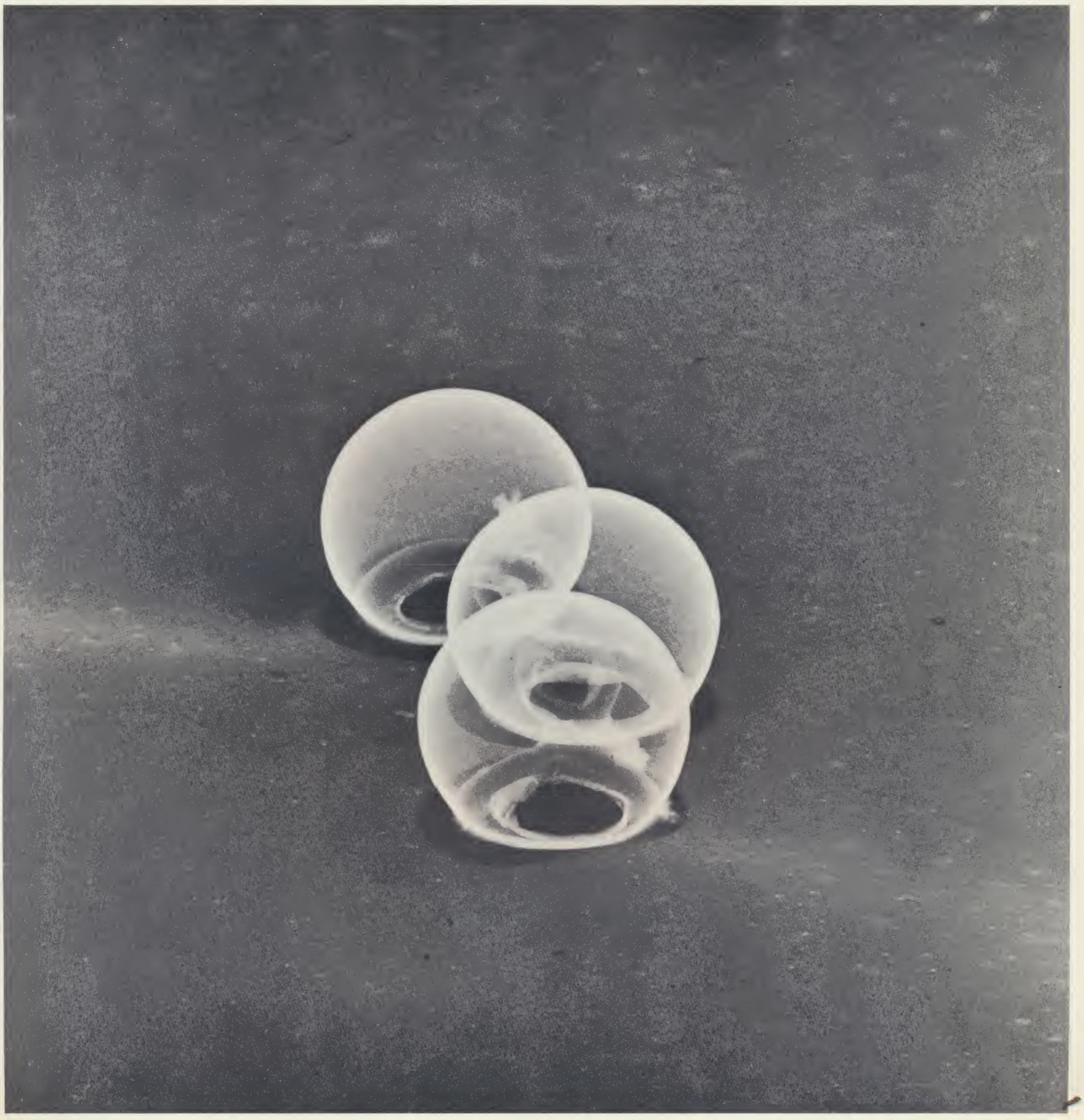
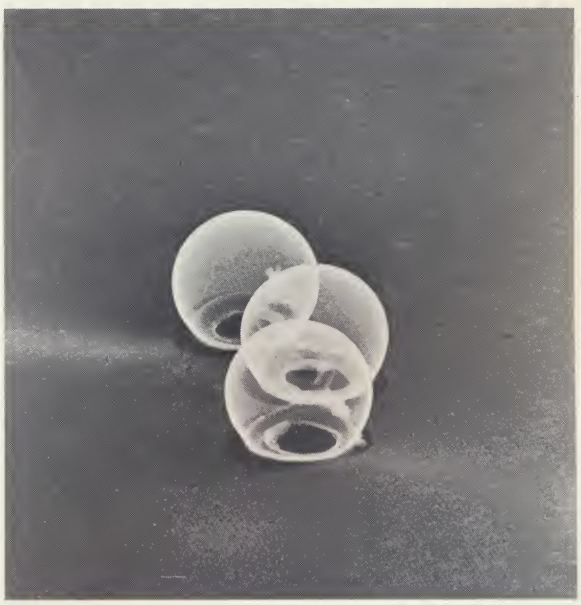
Scientific Instruments News

Volume 13 | No. 3 | September 1968





COVER ILLUSTRATION

A replica of three adjacent polystyrene-latex particles magnified 47,000×, this issue's cover illustration is provided by Walter S. Kay of duPont's Jackson Laboratories, Wilmington, Delaware.

To prepare it for electron microscopy, Mr. Kay placed the sample of polystyrene-latex particles on a nitrocellulose film supported on a specimen screen. After shadowing, at a low angle, with a platinum-palladium alloy, the sample was carbon covered to form a continuous film. Then, the nitrocellulose base was dissolved away with acetone and the polystyrene with an appropriate solvent. The result is a hollow cast of the original particles shaped in the metal and supported by the carbon. For this micrograph, the specimen screen was tilted 43° from the horizontal.

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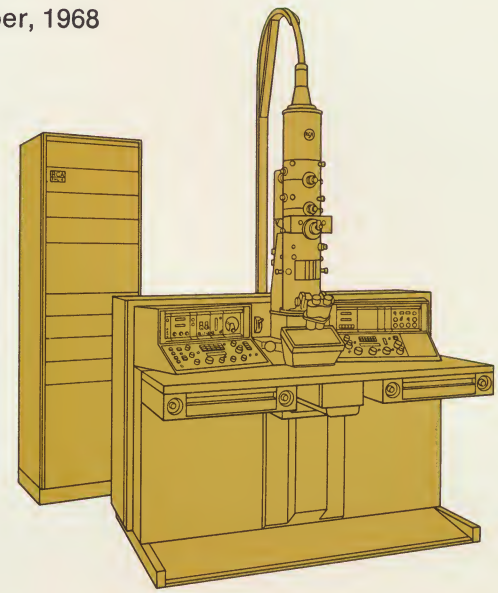

Scientific Instruments Marketing and Engineering
Building 15-4, Camden, New Jersey, U.S.A. 08102

Scientific Instruments News is published quarterly by RCA Scientific Instruments. This department is concerned with the design, manufacture and marketing of RCA Electron Microscopes, microscope accessories and associated products.

This publication is distributed to those engaged in the development and applications of electron microscopy—to provide a medium by which RCA-equipped microscopists may discuss their professional work.

The purposes of the publication are to disseminate technical information of professional value to the scientific community, by reporting on important developments in electron microscopy, revealing results of research by microscopists, and by describing various new products RCA offers.

Articles Solicited — Microscopists are invited to submit abstracts or manuscripts of articles suitable for publication. These should be original manuscripts, neither copyrighted nor pending publication elsewhere.

Send abstracts, manuscripts and associated material to the Editor, *Scientific Instruments News,* RCA, Building 15-5, Camden, N.J. 08102.

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News Briefs

ASCB TO MEET IN BOSTON

November 10 to 13, 1968, are the dates set aside for the Eighth Annual Meeting of the American Society for Cell Biology at the *Statler-Hilton* Hotel in Boston.

A commercial exhibition is scheduled as part of the meeting and RCA Scientific Instruments has been assigned Booth 15 at the "far" end of the exhibit area. We'll be receiving visitors at 8 A.M. on Monday, Tuesday and Wednesday (November 11-13).

The 1967 ASCB Meeting, held in Denver, attracted more than 1500 registrants. The Society anticipates even greater registration this year because the Meeting coincides with the annual meeting of the Genetics Society of America at the nearby *Sheraton-Boston* Hotel.

UNIV. OF MONTREAL EMU-4 ASSIGNED TO SYNAPSE STUDY

One of the EMU-4 electron microscopes serving science in Canada is installed in a newly-organized research center of the University of Montreal.

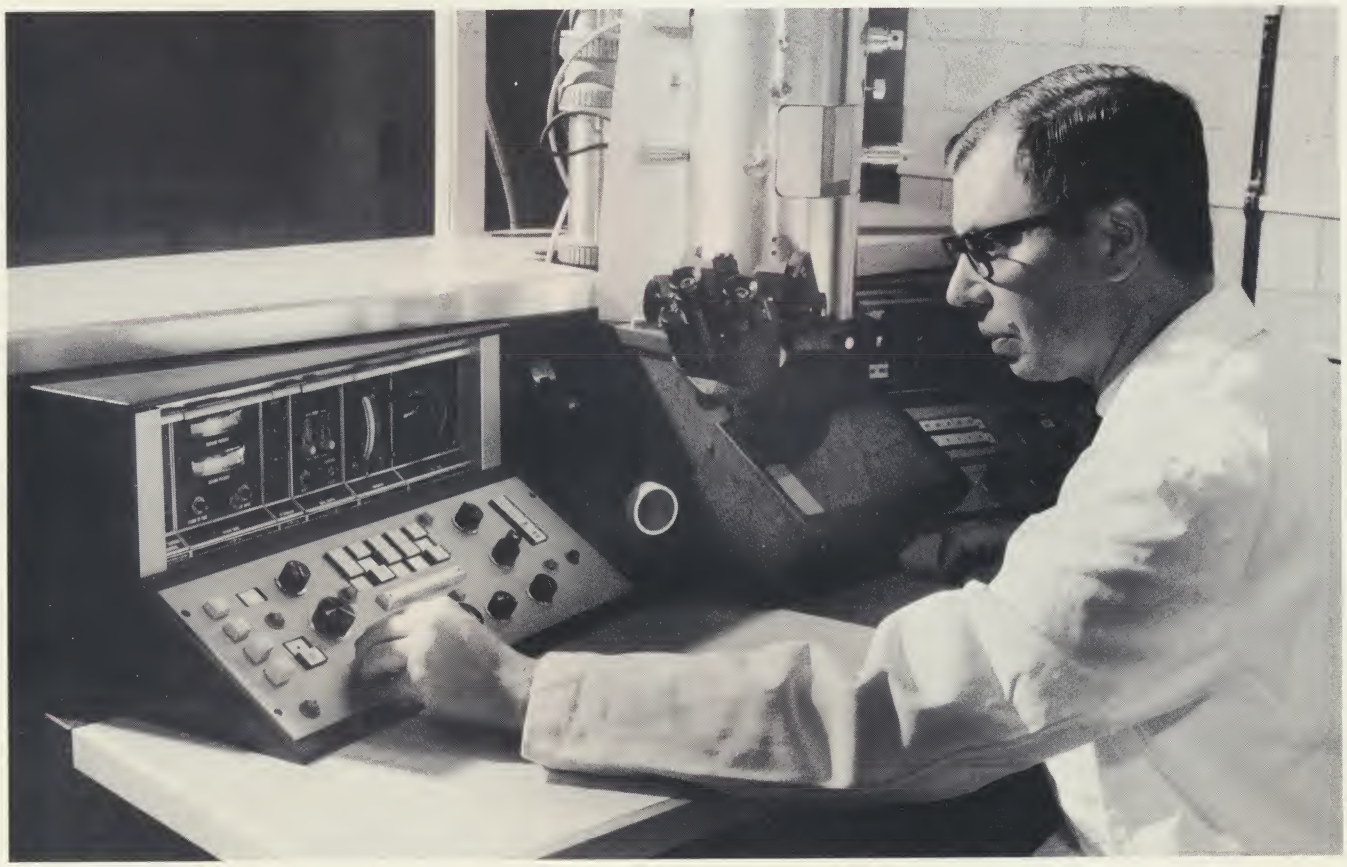
The Center is directed by Dr. Herbert H. Jasper, a founder of the *International Brain Research Organization,* now a professor of neurophysiology at the University of Montreal.

Dr. Marc Colonnier directs the use of the EMU-4. The instrument was financed with funds from Canada's Medical Research Council and U.S.A.'s National Science Foundation. The instrument was purchased via RCA Victor Co., Ltd., Montreal.

Dr. Colonnier, like Dr. Jasper, is a neurophysiologist. The group he heads consists of investigators Dr. J. P. Cordeau, Dr. Nico van Gelder, and Dr. Fernand Roberge. Dr. Cordeau is an engineer as well as chairman of the university's department of physiology; Dr. van Gelder is a neurochemist and Dr. Roberge, a biomedical engineer and assistant professor of physiology.

The group's research includes studies of what goes on at the synapse — the point at which a nervous impulse passes from one neuron to another.

Dr. Marc Colonnier at the console of the new EMU-4 installed at the University of Montreal.



RCA REPLACEMENT PARTS DEPOT AGAIN ENLARGED

Construction has started on a 150,000 square foot addition to RCA's three-year-old *Parts and Accessories* distribution center in Deptford, New Jersey, a Camden suburb. The addition will almost double the capacity of the almost-new plant.

Parts and Accessories is the replacement-parts function of RCA and it is this division's responsibility to inventory and market some 115,000 items of replacement parts for RCA electron microscopes and other products for as long as there is a need for such replacements.

It was only three years ago when the division moved to this new 200,000 square-foot plant from Camden. Paul B. Garver, vice president and general manager of the division said, "Our business has grown extensively, both in this country and abroad. The business increase, in fact, has outmoded in three years a facility we had expected to be adequate for 10 years."

An RCA computer does much of the bookwork of maintaining the vast inventories of replacement parts and accessories.

Air view of RCA Parts and Accessories' warehouse facility at Deptford, N.J., a suburb of Camden. A 150,000 square-foot addition, now under construction, will almost double the facility's floor area.

BASTE BASED IN ATLANTA

Robert D. Baste

Robert D. Baste is the new RCA Scientific Instruments Sales Representative for the southern United States. Mr. Baste reports to Neil Vander Dussen, RCA Scientific Instruments Marketing Manager.

As Regional Sales Representative, Mr. Baste is responsible for all RCA Scientific Instruments sales in the eleven states between the capes of North Carolina and the western panhandle of Texas, as far north as the Oklahoma/Kansas State Line and south to the Florida Keys. His base of operation is the RCA Building in Atlanta (see rear cover).

Bob Baste, born thirty years ago in Grove City, western Pennsylvania, earned his BS in Biology (1960) from Slippery Rock State College (Pa.) and continued his studies with graduate work at the University of Pittsburgh and West Virginia's Wesleyan University.

Bob gained his professional experience through a four-year relationship with Baxter Laboratories (Inc.) as a medical instrumentation sales representative and, most recently, two years as a sales engineer for the Packard Instrument Company.

Having now completed specialized training as an RCA electron microscope salesman, Bob looks forward to meeting all of the electron microscopists in the South, soon.

Bob, wife Carole Ann and their three sons, Scott Bradley (7), Robert Arthur (6) and Randy Allan (4) make their home in Stone Mountain, Georgia, a community in the hills some 20 miles east of Atlanta.

ERRATUM

The article "Cleaning Platinum and Molybdenum Apertures for Storage and Re-Use" (Vol. 12, No. 2, August, 1967), failed to point out that **molybdenum** aperture discs must be heated in the **reducing** portion of the flame. Heating the molybdenum discs in the **oxidizing** portion of the flame is apt to destroy the disc's usefulness.

THE ELECTRON MICROSCOPE IN CANCER RESEARCH

ROBERT E. BROOKS

Department of Pathology
University of Oregon Medical School
Portland, Oregon 97201

Abstract

The electron microscope has been used in cancer research for two decades. The hopes of many early microscopists to discover fundamental, meaningful ultrastructural differences between tumor cells and normal cells of origin have not been realized. Nevertheless, many opportunities for the electron microscopist in cancer research still exist. Detailed ultrastructural studies of carefully selected tumor systems may yet yield valuable insights into the neoplastic process. Cytochemical, immunochemical, and autoradiographic techniques, carried out at the electron microscope level, should find many applications in cancer research, particularly when applied in relation to new biochemical discoveries. Improvement in the analysis of chromosome fine structure would be invaluable in the study of cancer cells. If tumor systems of lower invertebrates become available, these may prove to be especially advantageous for electron microscopic study. In all cases, the electron microscopist in the field of cancer research will want to keep abreast of new ideas and theories concerning neoplasia.

Résumé

Le microscope électronique a été utilisé depuis deux décades lors de recherches sur le cancer. Les espoirs de nombreux microscopistes de découvrir les différences fondamentales, valables, ultrostructurales entre les cellules tumorales et les cellules normales sont restés jusqu'ici déçus. Cependant, il reste encore au microscopiste électronique de nombreuses possibilités dans la recherche sur le cancer. L'étude détaillée de l'ultrastructure de tumeurs soigneusement sélectionnées peut encore donner quelques aperçus valables du processus neoplastique. Les techniques cytochimiques, immunochimiques et autoradiographiques devraient trouver de nombreuses applications dans la recherche sur le cancer si elles sont utilisées au niveau du microscope électronique et particulièrement lorsqu'elles sont appliquées en relation avec les découvertes biochimiques récentes. L'amélioration de l'analyse de la structure fine des chromosomes serait de grande valeur dans l'étude des cellules cancéreuses. Si les systèmes tumoraux des invertébrés inférieurs étaient disponibles ils seraient spécialement utiles dans l'étude de la microscopie électronique. Dans tous les cas le microscopiste électronique dans le domaine de la recherche sur le cancer désirera toujours se tenir au courant des nouvelles idées et des nouvelles théories concernant la neoplasie.

When the electron microscope first came into general use for the study of tissues, there was hope that the great resolving power of this new instrument would reveal fundamental morphological differences between normal and cancerous cells. Now, twenty years after the first studies on the fine structure of neoplastic cells[1], it would appear that the original hope has not been realized.

Several reviewers have analyzed the findings in this research field and have arrived at the same conclusion. Dalton and Felix[2], in an early review article, stated, "Thus, in regard to the detailed structure of normally occurring components of the nucleus and cytoplasm, we have found no unequivocal evidence of qualitative differences between normal and malignant cells." Several years later, Luse[3] wrote in a review article, "The increased resolution of electron microscopy has failed to reveal any characteristics peculiar to neoplastic cells and absent from all normal cells." In a similar vein, Bernhard[4] stated in a 1963 review article, "To date, it has been impossible to link the malignant process to specific fine structural alterations, and considering methodological limitations one may even wonder if this will ever be possible." This outlook was restated in a recent atlas of the human lymph node by Bernhard and Leplus[5] where they wrote, "The electron microscope can certainly show better than the light microscope the classical cytological features of the cancer cell, but these changes may be absent and are not really specific for the neoplastic process. It would seem today that there is very little hope of detecting the initial stage of malignant degeneration by means of electron microscopical techniques."

It does not follow, however, that the electron microscope has no role to play in cancer research. The writers quoted above have emphasized this very point. The electron microscope can be and is being used to elucidate many aspects of the cancer problem. It is particularly useful in searching for and characterizing viruses in tumors. It can be and is used in long-term studies of experimental tumor systems. It is and will be of increasing value in the histochemical, autoradiographic, and immunochemical examination of preneoplastic and neoplastic tissues. Moreover, as new concepts arise from biochemical and other basic investigations, it would be expected that new avenues for electron microscopic exploration will become evident. Therefore, although the strictly morphological studies of tumors (especially single cases of human cancer) has, so far, offered few new insights, there still remain many opportunities for the electron microscopist in cancer research. Mention of a few such opportunities will be given in what follows.

The careful electron microscopic study of inducible tumors at various stages from the normal tissue to the full-blown tumor may be profitable in certain cases. There are known to be serious difficulties, however, in such an undertaking. Only a few cells out of very many exposed to any particular oncogen become cancerous. The chances of finding and recognizing significant morphological changes in the few positively effected cells will be slight. Nevertheless, accurate descrip-

tions of early ultrastructural changes in cells exposed to oncogens are badly needed, and the amount of work involved in this type of search is, perhaps, justified.

Of the newer methods which electron microscopists have utilized for identifying subcellular components, cytochemistry holds much promise. It is recognized that extensive study by many investigators has failed to demonstrate key biochemical steps in the transformation of normal cells to tumor cells. It is further recognized that existent cytochemical methods deal only with a very few of the extremely numerous biochemical entities. Nevertheless, continued biochemical research will unquestionably provide new data which will give rise to new hypotheses. Because some of the hypotheses will be testable by cytochemical techniques, it is expected that electron microscopists will use existent techniques, where applicable, and will want to devise new ones as they become needed.

Because viruses are implicated as the etiological agent in several animal tumors, and as the electron microscope is the most useful tool for the morphological study of viruses, there has been an understandable tendency to search all obtainable tumors, including human tumors, for the presence of virus particles. This approach, while providing valuable data when positive findings are made, does not exclude the existence of virus in tumors when the findings are negative, and has not yet provided significant information as to the relation of the virus particles to the neoplastic process. It may be suggested that the autoradiographic method could provide useful information in this area. For example, the ultrastructure of cells treated with radioactively labeled oncogenic viruses could be compared with that of cells treated with radioactively labeled infectious virus. An analysis of the ultrastructural differences found may reveal clues concerning the unique action of the oncogenic viruses.

Another use of autoradiography at the electron microscope level, would be in the study of the subcellular localization of labeled chemical oncogens. The oncogens could be administered to whole animals, or where detoxification is a consideration, to cell or tissue cultures.

Immunochemical techniques suitable for electron microscopy are slowly being developed. Such techniques should be used to test the concepts which link immunology and cancer.

Virtually untapped, but potentially valuable as experimental animals, are the lower invertebrates. Because of their small size, these animals are especially advantageous to the electron microscopist. The relatively few cell numbers, cell types, and tissue forms should make the detection and study of neoplastic changes much easier at the electron microscope level.

Crude techniques for studying whole chromosomes have been developed, and it is expected that more refined techniques, useful in detecting subgross abnormalities, will be discovered. Inasmuch as many current theories of oncogenesis implicate chromosomal changes, the examination of neoplastic cell chromosomes would certainly seem to be indicated.

It may be predicted that some of the techniques mentioned above, as well as others, will provide tools to keep the electron microscopist actively engaged in cancer research. However, as in any field of research, tools are most valuable when employed in the service of ideas. As Cecil Hall has written, ". . . Experimentation is neither infallible nor productive except when guided by theoretical reasoning." There are many ideas and theories concerning cancer. The better theories generate questions. Some of these questions may be answered by ultrastructural data. The electron microscopist who is aware of the questions will apply his special research skills most profitably.

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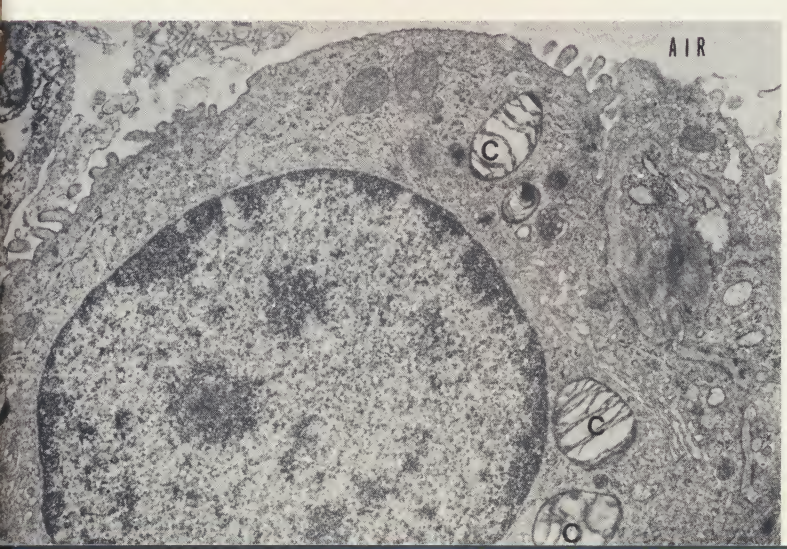
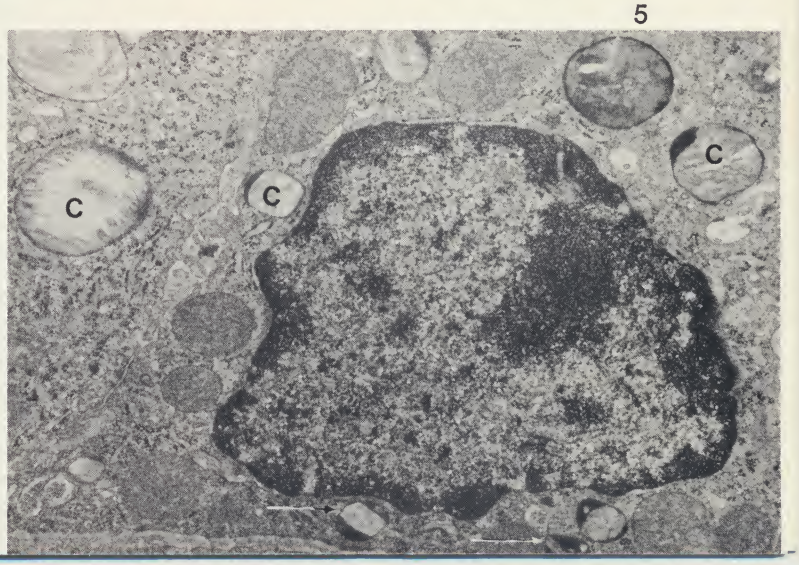
1. Porter, K. R. and Thompson, H. P. *Some morphological features of cultured rat sarcoma cells as revealed by the electron microscope.* Canc. Res. 7:431-38, 1947.
2. Dalton, A. J. and Felix, M. D. *The electron microscopy of normal and malignant cells.* Ann. N. Y. Acad. Sci. *63*: 1117-40, 1956.
3. Luse, S. *Ultrastructural characteristics of normal and neoplastic cells. In:* Progress in Experimental Tumor Research, F. Homburger (Ed.). Vol. II, pp. 1-35. Basel/New York, S. Karger, 1961.
4. Bernhard, W. *Some problems of fine structure in tumor cells. In:* Progress in Experimental Tumor Research, F. Homburger (Ed.). Vol. III, pp. 1-34. Basel/New York, S. Karger, 1963.
5. Bernhard, W. and Leplus, R. *Fine structure of the normal and malignant human lymph node.* Oxford, Pergamon Press, 1964.

Fig. 1. A type B alveolar cell from the lung of an oncogen (urethane) treated, tumor-bearing, mouse. This normal-appearing cell is of a type from which the common mouse-lung adenoma arises. The cell is characterized by structurally unique, lamelli-containing, cytosomes (marked with "C"). This cell, and the tumor cell shown in Fig. 2, is from lung tissue fixed with glutaraldehyde-paraformaldehyde and postfixed with osmium tetroxide solutions. ×13,000.

Fig. 2. An adenoma cell from the lung of the same mouse. The two cells are morphologically very similar, reflecting the well differentiated, benign state of the mouse-lung tumor. One ultrastructural difference consistently found relates to the cytosomes. Some tumor cell cytosomes (arrows at lower edge) contain, in place of the usual widely spaced lamellar material, a regularly-shaped interior compartment containing closely spaced lamellar material. In some instances, depending upon the fixative used, this cytosomal compartment contains highly ordered, crystalline material. Detailed, fine-structure comparison of normal and tumor cells may be profitable in some instances. This form of study represents the more "classical" approach taken by electron microscopists and will be pursued in selected tumor systems. ×13,000.





ELECTRON MICROSCOPIC TECHNIQUES FOR SHOWING EMBRITTLEMENT IN 18Ni MARAGING STEEL

D. A. GOMRICK and G. J. SPAEDER

U. S. Steel Corporation, Applied Research Laboratory, Monroeville, Pennsylvania.

Abstract

Embrittlement in 18Ni maraging steel was revealed by use of a two-step electron metallographic procedure. Examination of carbon-extraction fractographs of embrittled samples showed that they contained an inordinately large amount of a precipitate phase, indentified by electron diffraction as titanium carbonitride. Examination of carbon-extraction replicas of polished and etched cross sections of the embrittled steel showed that the precipitate was located in prior austenite grain boundaries, a fact that indicates that the fracture path was along these boundaries. Thus, the combination of the two procedures revealed the presence of a second phase that is responsible for the embrittlement and also defines the fracture path.

Résumé

La fragilité de l'acier 18Ni vieilli à la martensite a été mise en évidence par un procédé de métallographie électronique à deux échelons. L'examen de fractogrammes d'extraction du carbone dans des échantillons ainsi rendus fragiles a indiqué qu'ils contenaient une quantité excessive d'une phase de précipité que la diffraction électronique a révélé consister en carbonitrure de titane. L'examen de doubles des extraits de carbone dans des coupes transversales de l'acier fragile, polies et attaquées, a indiqué que le précipité se trouvait sur les lignes de démarcation précédentes des grains d'austénite, ce qui montre que la ligne de rupture suit ces lignes de démarcation. La combinaison des deux procédés a révélé ainsi la présence d'une deuxième phase qui entraîne la fragilité et qui détermine également la ligne de rupture.

Over the past several years, it has been well documented that certain thermal treatments result in moderate-to-severe embrittlement in 18Ni maraging steel. In general, the microstructural changes that are responsible for this embrittlement cannot be observed with the light microscope. However, a sequence of two, fairly simple, metallographic procedures—each involving the use of the electron microscope—provides an explanation for the embrittlement. The first procedure involves an examination at high magnifications of the fracture-surface of embrittled and unembrittled samples, and the second procedure involves a similar examination of a polished and etched surface. The combination of the two procedures defines the fracture path and also reveals the presence of a second phase that is responsible for the embrittlement.

Specimen Preparation

One-step, direct-carbon replicas of the fracture surfaces of embrittled and unembrittled samples were obtained from freshly-broken Charpy V-notch impact specimens. After a representative area of the fracture surface was selected for

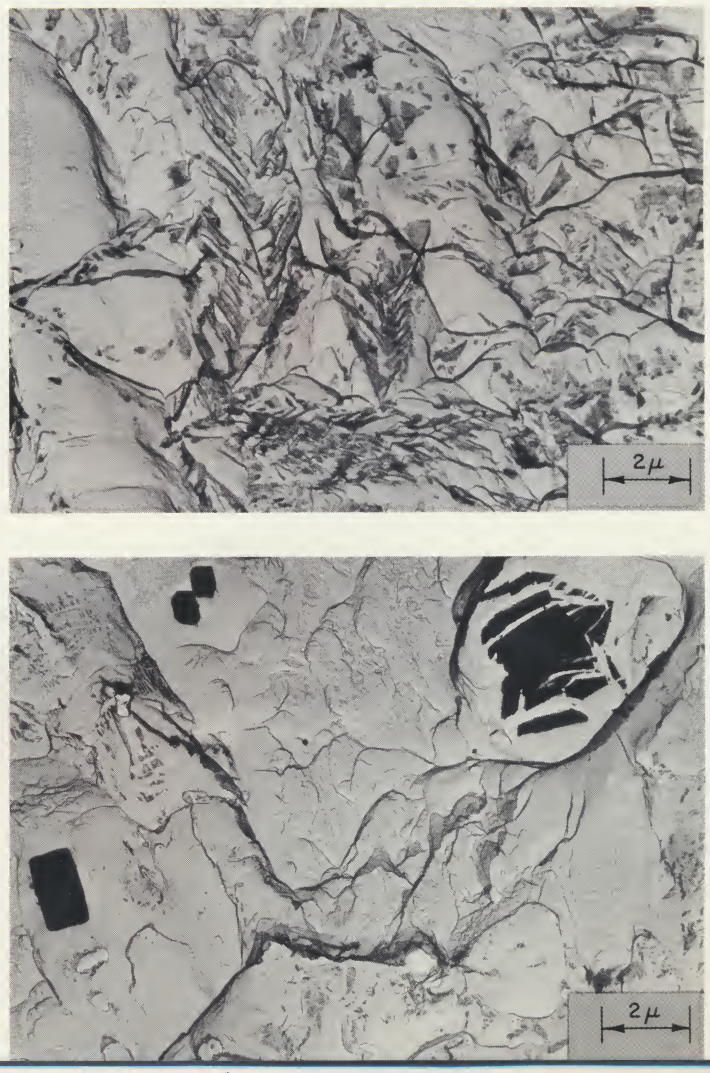


Fig. 1. Electron micrographs of extraction fractograph of embrittled 18Ni maraging steel.
(ASM Trans. Quart., Vol. 60, No. 3, 1967, p. 422)

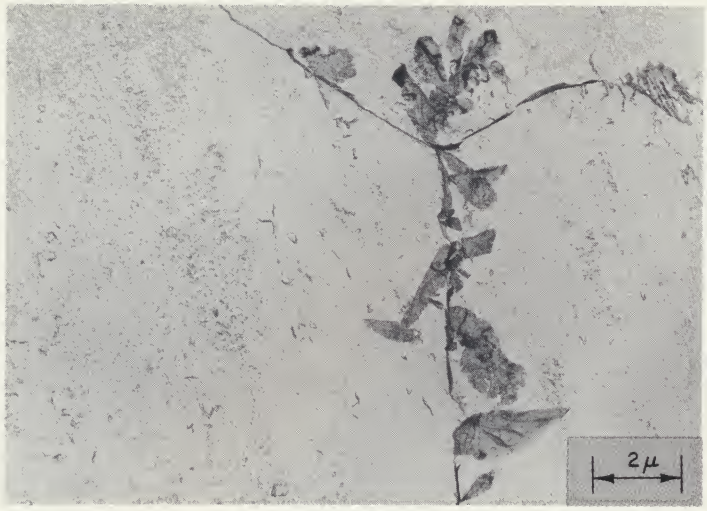
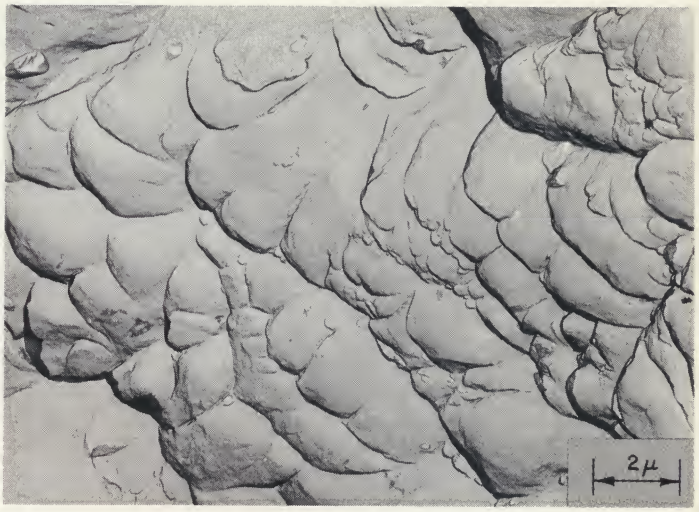


Fig. 2. Electron micrographs of carbon-extraction replica of polished and etched surface of embrittled 18Ni maraging steel.
(ASM Trans. Quart., Vol. 60, No. 3, p. 423)



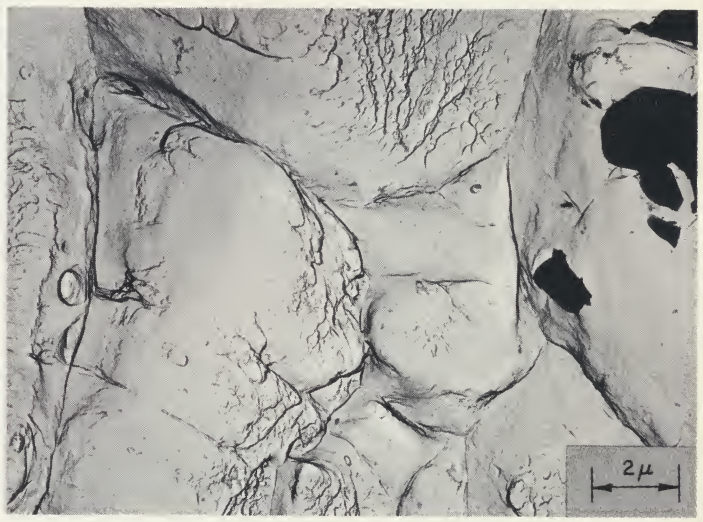


Fig. 3. Electron micrographs of extraction fractograph of unembrittled 18Ni maraging steel.
(ASM Trans. Quart., Vol. 60, No. 3, p. 424)

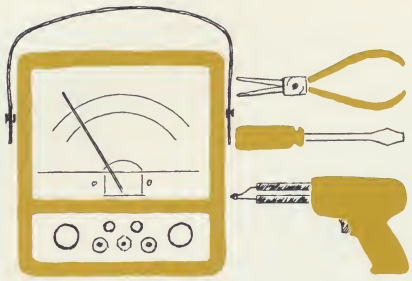
examination, the surrounding area was masked with a "stop-off" lacquer to allow carbon deposition only on the area of interest. The specimen was then placed 6 inches (152 mm) below a pair of carbon rods in an evaporator capable of reducing the pressure to about 10^{-5} mm of mercury. At such low pressures, a film of carbon about 300 angstroms thick can be deposited on the specimen in 9 to 10 seconds by resistance heating at the contact point of the two carbon rods. To remove the carbon film from the fracture surface, the area covered by the carbon was first scored with a scriber into a convenient size for viewing on the electron microscope. The specimen was then placed in a solution consisting of 100 ml of methyl alcohol, 10 ml of nitric acid and 5 ml of hydrochloric acid. This solution dissolves the metal below the carbon film and thereby frees the film from the fracture surface. After about two hours in the solution (when the edges of the carbon squares were free from the surface), the specimen was removed carefully from the acid bath and placed in methyl alcohol. Gentle agitation in the alcohol caused the carbon film, along with any insoluble precipitated particles, to break loose from the fracture surface. The film was then carefully placed on a 200-mesh specimen screen and rinsed thoroughly in methyl alcohol. At this point the carbon film, now a replica of the fracture surface, was ready for viewing in the electron microscope.

The procedure used for the preparation of carbon-extraction replicas of the polished and etched surfaces of embrittled and unembrittled specimens was identical with that used for the preparation of replicas of the fracture surfaces except that the carbon was evaporated on a polished and lightly etched (2% nital) section of the material.

Results of Metallographic Study

The replicas of the fracture surfaces and of the polished and etched surfaces were examined in the electron microscope and photographed at a magnification of ×4000. Figs. 1 and 2 are micrographs of the fracture surface and of the polished and etched surface, respectively, of the embrittled steel. Figs. 3 and 4 are similar micrographs of an unembrittled sample. Figs. 1A and 1B show that two types of precipitate were present on the fracture sur-

(continued on page 9)



MEET THE SERVICE ENGINEER

JOE REIBEISEN

Based in the Northeast Field Office (see rear cover) of RCA Service Co. is a serious, dedicated and mature microscope-service engineer who is one of those hard-to-find native New Yorkers. He was born in the heart of Manhattan some 60 years ago and grew up in Brooklyn, just across the East River.

Joe Reibeisen took on his basic education in the public schools of New York City and earned a BS in electrical engineering from Cooper Union Institute of Technology during 1927. He is a Licensed Professional Engineer in the State of New York.

After graduation, Joe went to work for the City as a civil engineer for New York's then expanding subway system. During the next two years, he had something to do with the construction of many of the lines which transport millions daily.

In 1929, Mr. Reibeisen joined Electrical Research Products, Inc. as a sound (audio) engineer. In this capacity, he installed motion-picture-sound equipment in many of the dozens of midwestern theaters equipping themselves for the coming of the sound track. This led him to his first position, in 1930, with RCA as a member of the *Photophone* Division, the organization responsible for theater equipment sales and service.

Three years later, in 1933, Joe decided it was time for a business of his own, and he formed Standard Sound Service, an organization offering theater-sound service to some 80 theaters in Brooklyn and Long Island. After five years of being his own boss, Joe returned to RCA, this time installing large-scale music-paging sound systems in industrial complexes. The one he remembers most vividly is the vast system in the Brooklyn Navy Yard.

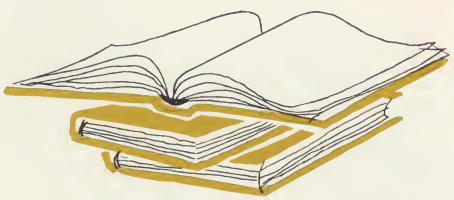
In 1945, he became an instructor at the RCA Signal Corps School in Philadelphia. As a member of the faculty he lectured the theory and operation of Airborne Radar Altimeters. This, in turn, led to a position as a representative of RCA Service Co. to the White House, the President's Cabinet and several commercial accounts in the Washington (D.C.) area.

In 1948, Joe became a member of a group assigned the task of demonstrating television – particularly the RCA system – to prospective broadcasters. This led, quite naturally, to closed-circuit television work and he figured in the installation of the first commercially-built, large-screen television system to be installed in Brooklyn's (N.Y.) Fox Theater. As if seeking to top that achievement, he followed it by contributing to the first large-screen, *color*-TV installation which went into NBC's color studios in Brooklyn. The screen size measured 15 x 20 *feet*.

In 1958, Joe Reibeisen transferred to Industrial and Scientific Service where he became an electron-microscope service engineer. He is proficient in the intricacies of every RCA electron microscope model from the EMB to the EMU-4. (The EMB was first made during 1941.) In all, he services some 25 RCA instruments in New York, New Jersey and Connecticut.

Joe and Anne Reibeisen make their home in the Flatbush section of Brooklyn where they share their hobbies of dogs, photography and collecting 18th-Century porcelain. Joe, for the fun of it, technically analyzes and charts the trends in the securities market.

Joe Reibeisen is a most capable, serious and thorough microscope service engineer. His clients consider themselves most fortunate to be the ones whose instruments he services.



Any of the literature described in this column is available from RCA Scientific Instruments Advertising, Building 15-5, Camden, N.J. 08102. Please request each piece by number.

NEW LITERATURE

EMU-4 Column Cross-Section Wall Chart—3J5215

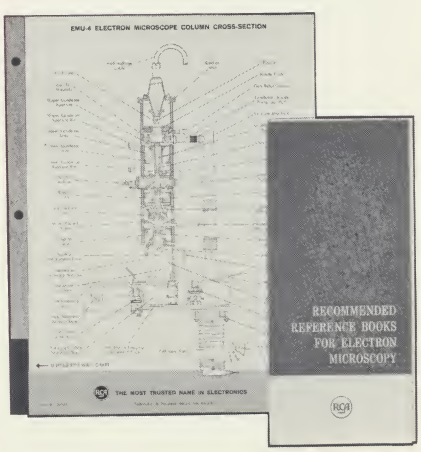
Designed to serve both as a wall chart and a notebook page, this 17- by 22-inch chart is printed on smooth-finished card stock and folded to a size compatible with the "standard" 3-hole looseleaf notebook.

The small chart is printed black-on-white however, the 17- by 22-inch chart is given a "reverse" treatment to increase its readability from a distance. The "call-outs" on the larger rendition are printed red-on-gray.

Recommended Reference Books for Electron Microscopy—Leaflet 3J5239

A pocket-sized leaflet which lists some 28 valuable reference works useful to electron microscopists or those interested in microscopy.

The works are arranged under four headings: "General Interest", "Physical Science", "Biological Science" and "Microscope Instrumentation" to aid in the selection of material most useful to the investigator's particular pursuit. Listings include date and place of publication.



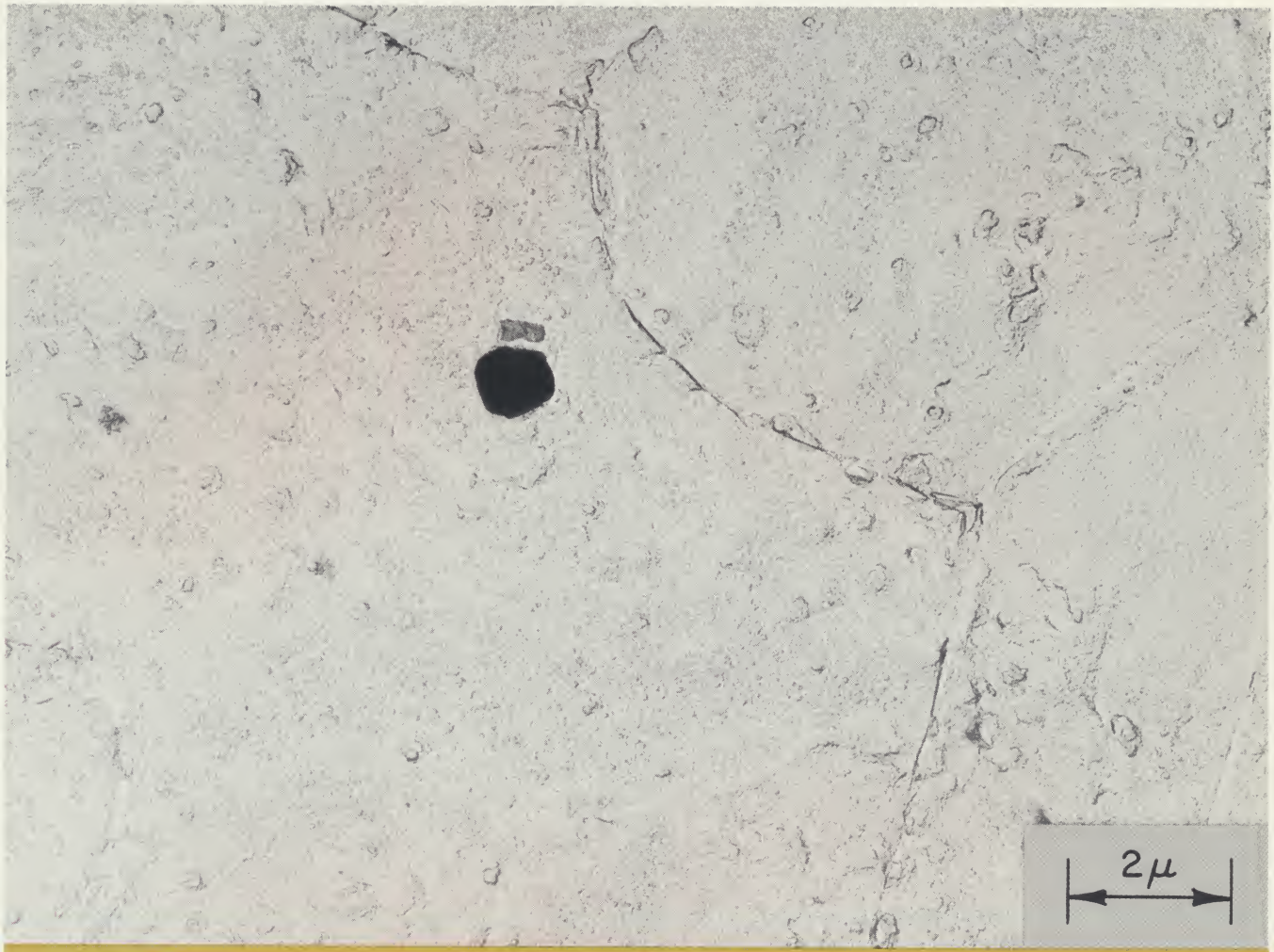


Fig. 4. Electron micrograph of carbon-extraction replica of polished and etched surface of unembrittled 18Ni maraging steel.
(ASM Trans. Quart., Vol. 60, No. 3, p. 424)

(continued from page 7)

face. One type, Fig. 1A, was feather-like in appearance and was probably a thin film at the prior austenite grain boundaries. The other type of precipitate was a dense, blocky particle, such as the particles shown in Fig. 1B.

Figs. 2A and 2B show that the film-type precipitate occurred primarily on the prior austenite grain boundaries, as evidenced by the fact that on the replicas of the polished and etched surfaces the areas near the grain boundaries were the only ones that contained as much precipitate as was observed on the fracture surface. This indicates that the fracture path was primarily along the prior austenite grain boundaries. Although not shown in the photomicrographs, the dense, blocky precipitate was observed in the matrix and was not preferentially associated with the prior austenite grain boundaries. The precipitate shown in Figs. 2A and 2B is probably a very thin film that was in a grain boundary prior to etching. The matrix material adjacent to the film was removed when the specimen was etched, leaving only the thin film from the boundary, which collapsed onto the surface of the specimen and thus appears (falsely) to extend out into the matrix from the boundary. Examination of the feather-like particles by selected-area diffraction indicated that this precipitate was primarily a titanium carbonitride Ti (C,N). The similarity of the *d*-spacings of TiC and TiN precludes positive identification of the particle as a nitride or carbide with this technique. A good diffraction pattern of the blocky precipitate could not be obtained because the particles were essentially opaque to the electron beam.

Figs. 3A, 3B, and 4 show that the feather-like precipitate observed on the replicas of the embrittled sample was almost nonexistent on the fracture surface or in the prior austenite grain boundaries of the unembrittled sample. The dense, blocky particles, however, were observed on the fracture surface and in the matrix, as illustrated in Figs. 3B and 4; the frequency of occurrence of the particles was about the same as that of the embrittled samples. Therefore, it was concluded that the feather-like grain-boundary precipitate is responsible for the observed embrittlement.

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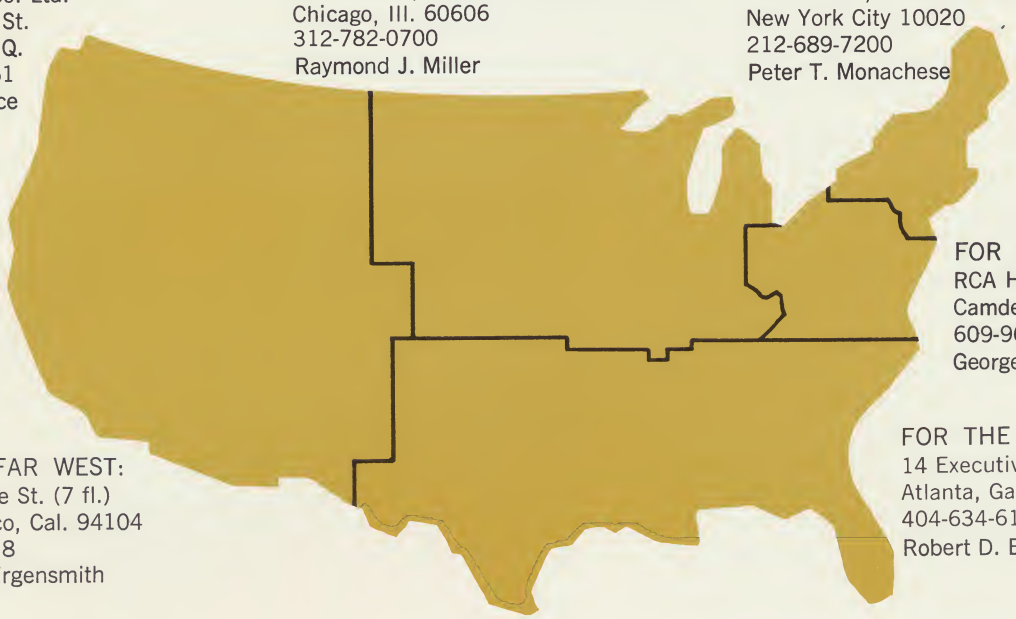

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